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Pattern of Apical Sensory Nerves in the Proboscis of *Macracanthorhynchus hirudinaceus* (Acanthocephala)

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ABSTRACT: The organization of the apical sensory organ in *Macracanthorhynchus hirudinaceus* includes a central core consisting of apical sensory nerves and a sensory support cell duct. Both nerves and duct terminate anteriorly in a pit or crateriform depression. About 30 μm posterior to the pit floor, the apical sensory nerves become quite pleomorphic. Simultaneously, they invade the sensory support cell duct. As the nerves move anteriorly, they branch repeatedly, whereas previously branched nerves are fusing. Some units remain on the outer surface of the sensory support cell duct while others remain enclosed by the duct. In either case, these combinations of nerves appear as thin sheets when viewed laterally. Those nerves on the outside periphery of the sensory support cell duct terminate near the apical pit and those enclosed by the duct become more nearly like thin cylinders that terminate in the walls of the pit.

KEY WORDS: Acanthocephala, nerves, proboscis, *Macracanthorhynchus hirudinaceus*.

The anterior terminus of the proboscis of *Macracanthorhynchus hirudinaceus* has a prominent cone-shaped elevation. This easily observed feature has generally been considered to serve in a sensory capacity. Dunagan and Miller (1983) reviewed the various terms that earlier authors had used in its description. Some of these terms such as "Tastpapille" suggest that this organ serves a chemosensory function. However, there is no experimental evidence to support this position. Nevertheless, circumstantial evidence, based on morphology, continues to suggest that this apical organ has a sensory role.

This study examines the relationship between apical sensory nerves and sensory support cell duct in the anterior terminus of the apical sensory organ of *M. hirudinaceus*.

Materials and Methods

Living worms were collected from pigs through the courtesy of Reelfoot Meat Packaging in Union City, Tennessee. Worms were transported to the laboratory in a Dewar flask containing intestinal contents. Detached specimens were rinsed in 30% seawater before fixing for 1 hr in a mixture of 2% glutaraldehyde and freshly prepared 2% formaldehyde in 0.1 M cacodylate buffer (pH 7.2) containing 2.0 mM EGTA and 1.0 mM $MgSO_4$. Specimens were then rinsed in 0.2 M cacodylate buffer at room temperature (RT) before being postfixed for 2 hr at RT in freshly prepared 1% OsO_4 and 1.5% $K_3Fe(CN)_6$. This procedure was followed by 3 20-min rinses in double-distilled water. Material was stained overnight in 1% aqueous uranyl acetate at 10°C. Specimens were dehydrated for 1–2 hr each in an ascending ethanol series (25, 50, 75, 95, 100%) followed by 3 20-min changes each in propylene oxide. The specimens were then infiltrated with increasing ratios (1:2, 1:1, 2:1) of propylene oxide : Spurr's epoxy resin over a 3-day period followed by 2 changes (1 day each) in pure Spurr's resin (1969). The specimens were then transferred into flat embedding molds, oriented as desired, and polymerized for 48 hr at 60°C. Serial sections were placed onto slot grids and examined in a Hitachi H500H transmission electron microscope operating at 50 kV.

Serial sections were cut at 80 nm thickness. Figures 5–42 depict every fifth section. Figure 4 is separated from Figure 5 by 560 nm and Figure 43 is separated from Figure 42 by 1.5 μm. The distance between Figure 4 and Figure 42 is 29.4 μm. The first section from each session of cutting was an orientation section of 1.4 μm in thickness.

Results

Posterior to the apical sensory core (Fig. 3), 2 anterior proboscis nerves, 2 sensory nerves, and a single sensory support cell duct are surrounded by muscles in the core of the proboscis. More anteriorly, this group encounters the apical sensory cone (ASC) where the anterior proboscis nerves divide and remain on the surface of the cone and the apical sensory nerves (SN) and sensory support cell duct (SSCD) enter the ASC (Fig. 2). One should note that while these last components are surrounded by the ASC they remain separated by 2 sets of membranes. In effect they are in a "hole" in the ASC. The SN and SSCD remain in the core of the ASC (Fig. 1) and eventually terminate in a pit on the apex of the proboscis. Approximately 30 μm (in the specimen shown) posterior to the pit, the well-defined SN and SSCD (Fig. 43) become progressively pleomorphic. The 2 nerves divide and redivide while becoming surrounded by the SSCD. This complex interaction is shown in Figures 4–43. (The

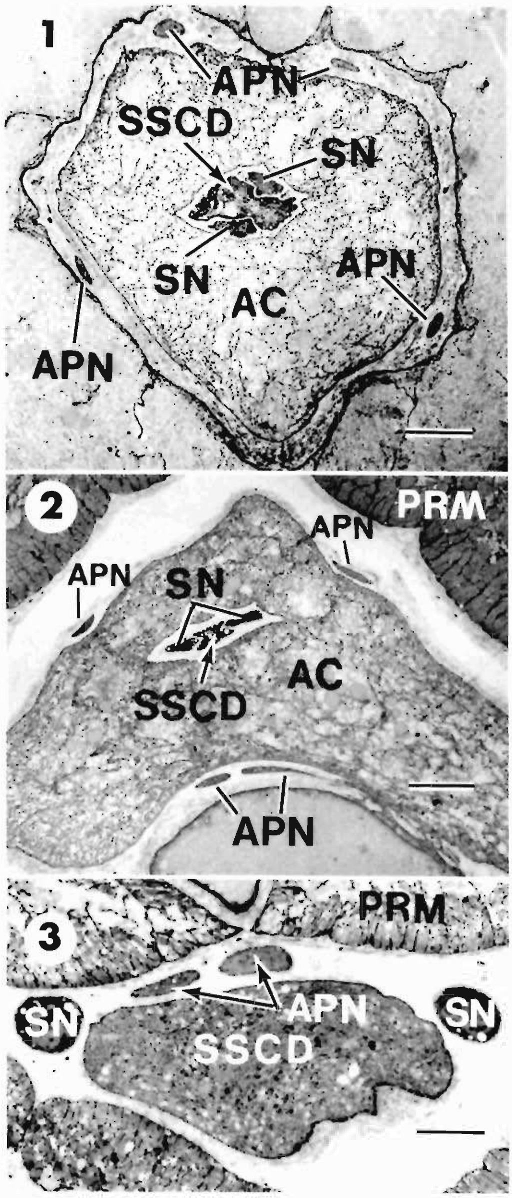
specimen is tilted slightly away from the viewer in the upper sections [Figs. 4–17] and results in an asymmetry in the pattern of nerves.) There is a constantly changing pattern in which the divided components unite only to divide once more. The individuality of the SN observed in Figure 43 is lost in this process. Eventually, they form discrete extensions (Figs. 5–13) going to the pit. Throughout this process there is close contact with the SSCD from the sensory support cell (stutzzelle) located adjacent to the cerebral ganglion.

Electron micrographs of different areas in the passage (Figs. 44–53) show the complexity of the events in this process. These pictures include the ASC through which the SN and SSCD are passing. Neither of the latter components reinvade the ASC in the process of passing through it. The ASC has not been included in the line drawings. Notice also that the SSCD has numerous fluid filled vesicles. These structures have not been included in the line drawings (Figs. 4–43).

Discussion

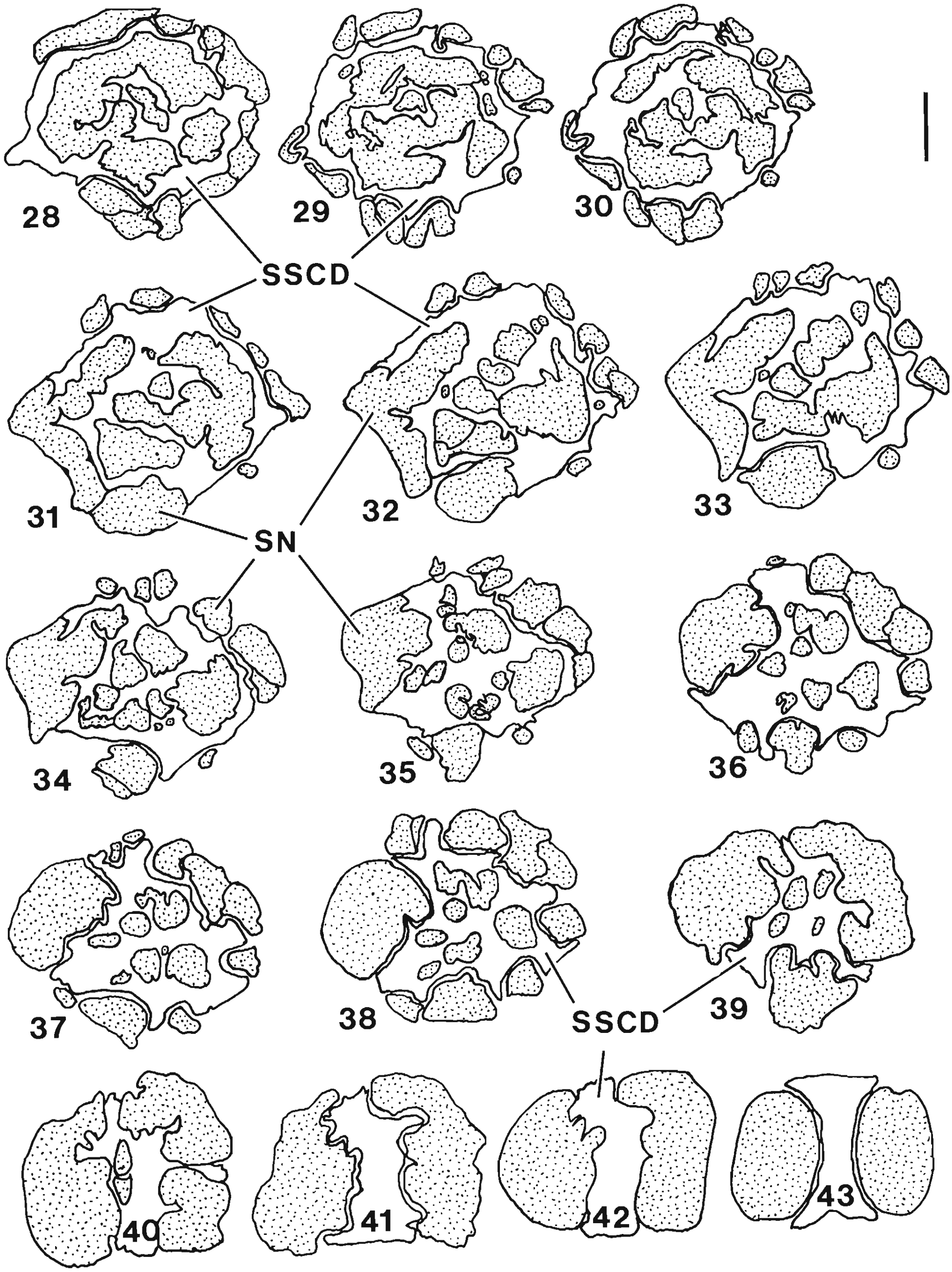
Hyman (1951) indicated that the area beneath the pit had a coiled nerve with a fusiform ending. Rauther (1930, p. 458) stated that 2 fibers of the medial nerve wind convolutedly upward and enter the apical pore forming a sensory papilla. Kaiser (1893, part 2, p. 3) stated that a nerve (probably sensory) from the anterior part of the cerebral ganglion traveled between the retractor muscles to the proboscis tip where it ended. Kaiser (1893, part 2, pp. 8–9) expanded on this earlier statement by adding that in the apical sensory cone "two nerve fibers from the anterior medial nerve twist themselves into a thick ball . . . which represents a sensory papilla." It appears that Kaiser interpreted this papilla as a mechanoreceptor related to the eversion of proboscis hooks. In 1876, Leuckart, according to Harada (1931), observed that the anterior medial nerve connected with the tactile papillae on the proboscis tip. Dunagan and Miller (1983) outlined the apical sense organ including the point of entry of the sensory nerves and the branch pattern of the anterior proboscis nerves. However, these authors were unable to resolve the anterior terminal organization of these nerves.

The figures presented herein depict an area of intense interaction of SN and SSCD in which the 2 original nerves branch many times throughout most of the anterior 30 μm of this system. Yet the branches do not remain separate but fuse in



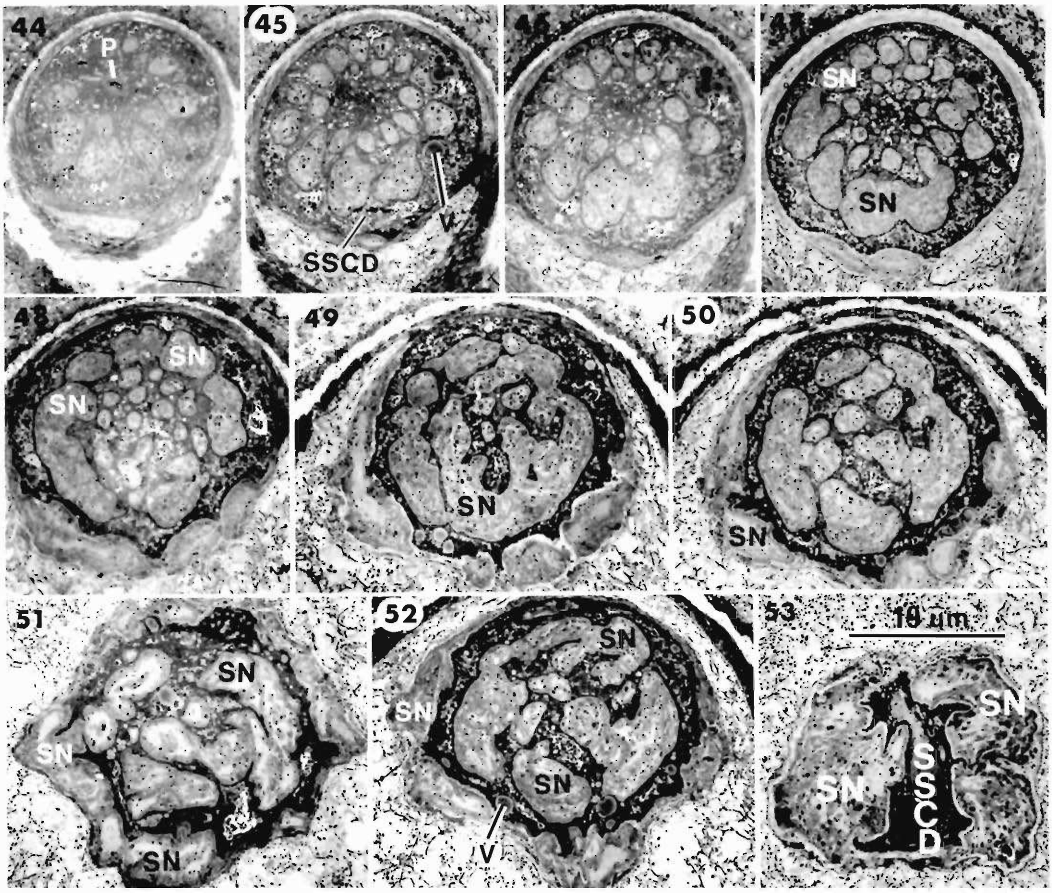
Figures 1–3. Nerves and sensory support cell duct in proboscis of *Macracanthorhynchus hirudinaceus*. Abbreviations: apical sensory nerve (SN); anterior proboscis nerve (APN); apical sensory cone (AC); proboscis retractor muscles (PRM); sensory support cell duct (SSCD). 1. Pattern of nerves posterior to apical sensory cone. Scale bar = 8 μm. 2. Pattern of nerves immediately anterior to their entry into apical sensory cone. Scale bar = 10 μm. 3. Pattern of nerves at midlength in apical sensory cone. Scale bar = 2.3 μm.

Figures 4–27. **Illustrated cross sections of sensory (stippled area) and sensory support cell duct at anterior terminus of proboscis of *Macracanthorhynchus hirudinaceus*. Abbreviations: apical sensory nerve (SN); sensory support cell duct (SSCD); apical pitlike opening on anterior of proboscis (P). Note that the surrounding tissue of the apical sensory cone (see electron micrographs) has been omitted. Scale bar = 9.8 μm.**



Figures 28–43. Illustrated cross sections of sensory nerves (stippled area) and sensory support cell duct at anterior terminus of proboscis of *Macracanthorhynchus hirudinaceus*. Abbreviations: apical sensory nerve (SN); sensory support cell duct (SSCD). Note that the surrounding tissue of the apical sensory cone (see electron micrographs) has been omitted. Scale bar = 9.8 μm.

unpredictable ways to form smaller numbers of larger units only to separate again into numerous branches near the pit of the anterior "papillae." We have completed 3 sets of serial sections on this area and the structural pattern (but not the exact points of branching) has remained the same. The failure of any of the SN components to interact with muscle suggests that these nerves have



Figures 44–53. Electron micrographs of core of apical sensory organ showing apical sensory cone enclosing apical sensory nerves and sensory support cell duct in proboscis of *Macracanthorhynchus hirudinaceus*. Abbreviations: apical sensory nerve (SN); apical pitlike opening on anterior of proboscis (P); sensory support cell duct (SSCD); vesicle (V). Scale bar = 19 μm.

nothing (directly) to do with hook movement or movement in other adjacent large muscle groups such as the proboscis retractors.

It is also clear that SN outside the SSCD terminate before the floor of the pit. The fact that some SN remain outside the SSCD and pit area suggests that the SN may be multifunctional.

Acknowledgments

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Literature Cited

Dunagan, T. T., and D. M. Miller. 1983. Apical sense organ of *Macracanthorhynchus hirudinaceus* (Acanthocephala). Journal of Parasitology 69:897–902.

Harada, I. 1931. Das Nervensystem von *Bolbosoma turbinella* (Dies.). Japanese Journal of Zoology 3: 161–199.

Hyman, H. L. 1951. The Invertebrates: Acanthocephala, Aschelminthes, and Entoprocta The Pseudocoelomate Bilateria. Vol. III. McGraw-Hill Book Company, New York, Toronto, and London. 572 pp.

Kaiser, J. E. 1893. Die Acanthocephalen und ihre Entwickelung. Pages 1–148 *in* R. Leuckart and C. Chun, eds. Bibliotheca Zoologica. *Zweiter Theil*, Heft 7. Theodor Fischer, Cassel.

Rauther, M. 1930. Sechste Klasse des Cladus Nemathelminthes Acanthocephala=Kratzwurmer. Pages 449–482 *in* W. Kukenthal und T. Krumbach, eds. Handbuch der Zoologie. 2 Band, 1 Halft. Walter De Gruyter & Company, Berlin and Leipzig.

Spurr, A. R. 1969. A low-viscosity epoxy resin embedding medium for electron microscopy. Journal of Ultrastructure Research 26:31.